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DEVELOPMENT OF INSECT RESISTANCE TO INSECTICIDES--II
A Critical Review of the Literature up to 1951

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In May 1949 the senior author (Babers 9) published a review covering the literature up to the fall of 1948 on the subject of the development of resistance by insects to insecticides. At that time only a few cases of the failure of DDT to control house flies in the field had been reported from this country, and the potential significance inherent in these failures was apparently not universally recognized. Recently, however, these failures have received great publicity in scientific publications and in the press.

In the earlier review it was noted that the following species had been reported as having developed resistance to insecticides: California red scale (Aonidiella aurantii (Mask.)), black scale (Saissetia oleae (Bern.)), citricola scale (Coccus pseudomagnoliarum (Kuw.)), codling moth (Carpocapsa pomonella (L.)), vinegar fly (Drosophila melanogaster Meig.), cotton aphid (Aphis gossypii Glov.), confused flour beetle (Tribolium confusum Duv.), blue tick (Boophilus decoloratus (Koch.)), citrus thrips (Scirtothrips citri (Moult.)), screw-worm (Callitroga americana (C. & P.)), an Italian subspecies of the northern house mosquito (Culex pipiens autogenicus), house fly (Musca domestica L.), and San Jose scale (Aspidiotus perniciosus Comst.).

It is the purpose of the present review to bring the subject of the development of insect resistance to insecticides up to date and to include a critical discussion of some of the causes of the various discrepancies in results reported by the several observers.

California Red Scale

Several papers overlooked in the earlier review have been noted. Busbey et al. (35) found that second-stage nymphs of the strain of red scale (Aonidiella aurantii (Mask.) resistant to hydrocyanic acid were more susceptible to trichloroacetonitrile than were the adult insects. The reverse is true when the insects are tested against hydrocyanic acid. The normal strain was not tested. Yust, Nelson, and Busbey (171) studied the effect of inbreeding on the susceptibility of the red scale to hydrocyanic acid. After several years no difference in susceptibility attributable to the inbreeding was noted.

Cressman (40) found large differences in the mortalities of scales

from various groves when the insects were sprayed in the field with comparable dosages of oil, or collected in the field and sprayed in the laboratory. Scales collected from groves in which extreme differences were noted and reared in the laboratory for several generations showed almost no differences in susceptibility when tested at intervals over a period of about 9 months. Cressman concluded that most of the observed variations in the susceptibility of field stocks of red scales to oil sprays were due to other than genetic differences. Whether or not any of the strains tested were those considered to be resistant to hydrocyanic acid was not stated. It is not clear how Cressman reached his conclusions concerning lack of genetic differences from the type and small amount of data presented. In the previous review (9) it was mentioned that the resistance of red scale to hydrocyanic acid had been shown by several investigators to be inherited. According to Lindgren and Dickson (91) there was no difference between the susceptibility of laboratory-reared stocks of the two strains to oil sprays. Cressman did not refer to this earlier work. Munger (119) compared the rate of development of red scales resistant to hydrocyanic acid with nonresistant insects and found no significant differences. He also (120) determined the effect of temperature on the biology of the two strains, and again no significant differences between the strains were noted.

Drosophila

Kalina (70) tried to develop a resistant strain of Drosophila melanogaster by rearing larvae produced from eggs laid by normal females in a medium containing 5 parts per million of DDT. Larval development was somewhat slower than normal but up to the time of early pupation no real difference was noted. In the later stages of pupation, development was abnormal. Adults usually failed to emerge, and those that did exhibited signs of DDT poisoning and soon died. The speculation was made that DDT was stored in the larval fat and was released during histolysis in the pupae. Whether or not experiments were performed using lower concentrations of DDT was not stated.

A strain of Drosophila resistant to DFDT (1,1,1-trichloro-2,2-bis (p-fluorophenyl)-ethane) was developed by Reimschneider and Rohrmann (137). After 32 generations of selection, resistance had developed so that an exposure to deposits of DFDT that caused 70 percent mortality in the normal strain caused 30 percent mortality in the resistant strain.

Resistance to carbon dioxide by Drosophila is apparently a normal condition, whereas susceptibility is abnormal. The sensitivity seems to depend on the presence in the cytoplasm of an agent called a genoid. L'Heritier and his associates (86,88,89,90) showed that the susceptibility to carbon dioxide could be transmitted by transplanting from susceptible to normal individuals ovaries, brain tissue, or imaginal discs of the eyes, legs or wings, or portions of the gut. Hemolymph as well as the centrifuged supernatant of crushed susceptible flies at any stage was shown to be infectious. Under certain conditions the exposure of sus-

ceptible flies to heat will "cure" their germ cells, and it is thus possible to obtain a genoid-free stock from any sensitive stock. They also found that X-rays would inactivate the genoid (87). This and further work by L'Heritier raised the question as to the possible similarity of the genoid to a virus, and L'Heritier and his collaborators are now approaching the subject by using techniques applicable to virus research. Such an approach will, if successful, throw light on the relation between viruses and genes and will have far reaching applications.

According to Goldstein (58, 59), if "normal" Drosophila males are made "sensitive" by the injection of sensitive hemolymph, they transmit the sensitivity to only a small percentage of their progeny, whereas if the males are normally sensitive a higher percentage of transmission occurs. It was found that the two types of males could be obtained from any stock. To explain this, it was suggested that the genoid of the synthetic sensitive stock had undergone a mutation limiting strongly its capacity to infect the male germ. Plus (133) determined the time required (incubation time) for Drosophila to become susceptible to carbon dioxide following injection with a suspension of genoids. If moderately high concentrations of genoids were used, a linear relation was observed between incubation time and the log of the number of infectious units injected. As an appendix to Plus's paper L'Heritier presents a mathematical treatment of these results. A review by L'Heritier (85) covers the subject of carbon dioxide sensitivity up to about 1947.

There is considerable variation in the susceptibility to ether among Drosophila virilis, D. americana, and D. texana. Crow (41) found that when the dosage was varied and the time of exposure held constant virilis was the most resistant, followed by americana and texana. These observed differences were small. When the dosage was constant and the time of exposure varied, virilis was the most resistant, then texana, and finally americana. These differences were large. Fox (53) prepared rabbit anti-sera from three mutant strains of lyophilized insects. By complement fixation tests, no differences in the anti-sera were noted. By precipitin ring tests, however, certain differences were noted. These differences were believed due not to the single major genic difference but to the multiple genic differences that must exist between such unrelated stocks.

Ticks

No reports of insecticide-resistant ticks in the United States have been noted. In sections of South Africa and to a lesser extent in Argentina, the problem is of considerable importance. Resistance to arsenic dips by the blue tick was first noted in 1940 in a small coastal area of South Africa. The resistance spread rapidly. It seemed to be inherited but other factors were definitely involved. Whitnall and Bradford (165) reported that if cattle infested with resistant ticks were transferred from the coastal area to an inland area, the ticks either disappeared because of their inability to survive in the new

environment or they succumbed to the normal arsenical dipping. Whitnall and Bradford in 1947 (165), and again in 1949 (166), and Thorburn (157) in 1947 reported that the resistant ticks were readily controlled with Gammexane dips. Whitnall and Bradford (166), also reported that the tick that caused sheep paralysis, Ixodes pilosus Koch, seemed somewhat resistant to the Gammexane but succumbed to the usual arsenic dips. Later in 1949 Whitnall and others (164, 167) and Haarer (60) reported that the arsenic-resistant tick had become resistant to gamma benzene hexachloride. In Argentina control of the blue tick with benzene hexachloride was still being obtained (51). Haarer (60) in discussing some of the economic aspects of the resistance of the blue tick stated, "On one ranch alone, 600 head of stock died in one year because of the failure of arsenic to control the blue tick. The first year Gammexane was used, the figure dropped to 70 head. It was no wonder the cattlemen were jubilant with the material and it is also easy to understand their perturbation at the fact that resistance to benzene hexachloride has now developed."

Red Spider Mite

Smith, Fulton, and Lung (150) reported excellent control of many greenhouse pests, including the two-spotted spider mite (Tetranychus bimaculatus Harvey), with parathion aerosols in 1948. Complete control of the same mite with parathion was also reported by Blauvelt and Hoffman (22) about the same time. They state "Parathion shows promise of being the most important material yet discovered for greenhouse pest control." Parathion was effective in some greenhouses for only about a year, when Smith and Fulton (147, 148, 149) and Garman (56) found that resistance to the insecticide had developed.

Garman (56) found that if resistant mites were transferred from greenhouses where they had infested roses to the laboratory where they were reared on bean plants, the resistance was lost in about 4 months. On the other hand, Smith and Fulton had reared resistant mites on beans for many months without exposure to insecticide and without any seeming loss of resistance. Garman found that the parathion resistance did not include resistance to tetraethyl pyrophosphate, p-chlorophenoxy methane, di(p-chlorophenyl) methyl carbinol, or "alkyl sulfite" (trade product of unstated chemical composition). Except for tetraethyl pyrophosphate, which is distantly related chemically to parathion, the other materials are not structurally related to that compound. Smith's colony, however, showed considerable resistance to the following other insecticides: Hexaethyl tetraphosphate, tetraethyl pyrophosphate, tetraethyl dithiopyrophosphate, 1,1-bis(p-chlorophenyl)ethanol, 2-(p-tert-butylphenoxy-1-methylethyl 2-chloroethyl sulfite, and p-chlorophenyl p-chlorobenzenesulfonate. Two of the above compounds were also tested by Garman, who reported his strain was not resistant to them. Fair control was still obtained by Smith using several of the above compounds although resistance was evident. Octamethyl pyrophosphoramide gave excellent control of resistant mites but whether or not there was any resistance to the compound was not determined.

In two greenhouse ranges in which Smith found "parathion-resistant" spider mites, resistance to hexaethyl tetraphosphate had been noted in 1947 when it was first used as an insecticide. Repeated applications and heavy dosages had, however, maintained control. The development of resistance then could not be attributed to the use of either hexaethyl tetraphosphate or parathion. The history of these greenhouse ranges prior to the first use of hexaethyl tetraphosphate is not known, but undoubtedly the usual insecticidal practices for the control of greenhouse pests had been followed.

E. W. Baker of this Bureau's Division of Insect Identification (personal communication) has been unable to find any morphological differences between the susceptible and resistant strains.

Palm (126) reports that the red spider mite in New York State developed resistance to ammonium potassium selenosulfide when this compound was used several years ago, and that cases of resistance to parathion are now occurring.

Neiswander and Morris (121) after a period in which excellent control of two-spotted spider mites on roses grown in a nutrient solution containing selenium was obtained, reported that a severe infestation developed on the plants that had received a high dose of selenium. At that time, the infestation was not attributed to resistance. Recently, however, Neiswander, Rodriguez, and Neiswander (122) have decided that the insects had become more resistant to the insecticide. They further conclude that wide variations between populations of the mite exist. The rate of development as well as the resistance to acaricides was influenced by the host plant. A mite population reared on roses is generally more resistant than one reared on beans. A population subjected to an acaricide for three or more generations may develop a partial immunity to that material. Neiswander et al. (122) state, "It appears that such a heterogeneous organism as the two-spotted spider mite may require that acaricidal materials be changed at frequent intervals in order to insure effective control."

Mosquitoes

Salt-marsh areas along the east coast of Florida have been treated with oil solutions of DDT at regular intervals since 1945. Before this period of treatment salt-marsh mosquitoes occurred in such numbers as to prevent full development of the area. After the treatment, freedom from mosquitoes was considered one of the shining examples of modern insect control measures. Recently the two most prolific insects involved, Aedes sollicitans (Wlkr.) and A. taeniorhynchus Wied., have been reported as developing resistance to DDT (17, 27, 37, 47, 48, 73, 106).

Deonier and his associates (47, 48) found that the DDT resistance was also apparent in the larvae. In addition to DDT the authors tested other insecticides--TDE, toxaphene, lindane, technical benzene hexachloride, and chlordane. They conclude, "Like DDT, TDE was much less effective against the adults from the treated area than it was against those from untreated areas, an indication that the adults were about

equally resistant to TDE and DDT. At the lower concentration (0.5 percent) the other materials also showed less toxicity to the mosquitoes from the treated area, but at the higher concentration (2 percent) this difference was not in evidence except possibly with toxaphene." The authors do not comment on the statistical significance of their data. In California, DDT has been reported as failing to control larvae of Culex spp. (72) and of Aedes nigromaculis (24).

Fay, Baker, and Grainger (50) attempted to develop resistance by exposing the common malaria mosquito (Anopheles quadrimaculatus Say) to low concentrations of insecticides in the laboratory. After one generation, some resistance had apparently been developed, but after several succeeding generations the resistance had not appreciably increased and the results did not seem to be conclusive. The experiments were then abandoned. The Malaria Eradication Program of the U. S. Public Health Service (26) has recently completed 5 years' operational work. Thousands of houses have been sprayed in the southeastern United States in an attempt to eradicate malaria by control of the mosquito vector. During this 5-year program, apparently no resistance to DDT has been evident. During the course of each season, however, there was a gradual decrease in the effectiveness of residual DDT. No indication as to the reason for this decrease was given but it is probable that loss of effectiveness was due to the normal aging of the deposited DDT. Mosna (114, 115) has reported that the DDT-resistant mosquito, Culex pipiens autogenicus (113, 118), was effectively controlled with Octa-klor (chlordane) and hexachlorocyclohexane (gamma benzene hexachloride). Only control dosages of these chemicals were used and no effort was made to determine whether any resistance to them was apparent. Mosna noted that in DDT-treated houses, although there was an increase in the number of live insects, there was also an increase in the number of dead ones. The resistant insects were able to transmit the resistance through the eight generations tested without exposure to DDT. According to Verolini (161) the adults of C. pipiens from Latina were resistant to DDT, but the larvae showed high mortality from exposure to DDT. No comparison was made to determine whether or not any resistance was present. Starting a laboratory colony of C. fatigans Wied. from a single egg mass, Newman, Aziz, and Koshi (124) found that successive generations showed greatly increased resistance to DDT and gammexane. The authors suggested that the increased resistance might be due to a rhythmic variation in the resistance of C. fatigans.

Gahan and others (54) compared the susceptibility of Anopheles psuedopunctipennis Theob. from areas in Mexico that had received four annual treatments with DDT to the susceptibility of mosquitoes from untreated areas. No difference was noted.

Knipling (80) reported that there is no evidence that mosquitoes in the Pacific Northwest have developed resistance although resistant strains have been suspected in Oregon.

House Flies

Development of resistance in the field and in the laboratory

House flies that have developed resistance to insecticides have been collected in the field in Italy (110), Greece (61), Egypt (55), Venezuela (160), Sweden (168), Denmark (71), England (65), Sicily (4 Peru (141), Mexico (160), and the United States (12). Strains with acquired resistance to various insecticides have been developed in several laboratories in the United States (15, 23, 29, 94, 103, 134), Sicily (42), and Great Britain (65). Busvine (36) collected several strains of Musca vicina Macq., the house fly of Ceylon, and found no resistance although the area had been sprayed with DDT for 2 years. Surveys have been made in several sections of this country (12, 30, 33, 64, 72, 74, 81, 99, 126, 134a, 139, 160) and in Italy (142, 143), and many strains of house flies resistant to insecticides found. Indeed, Upholt (160) states that his coworkers were unable to find a nonresistant strain in this country and contiguous areas of Mexico in 1949.

La Face (83), in a study of the biology of the fly and its importance as a vector of various diseases, discussed resistance to insecticides. DDT-resistant flies--Musca domestica and Stomoxys calcitrans (L.)--in Sweden were discussed by Kjellander (75, 76). No report of insecticide resistance in Russia has been noted. However, Rubtsov (14(after a visit to Missirolis' laboratory in Italy, summarized Missiroli's results and speculated somewhat as to the probable causes for the appearance of resistant insects.

Morrison and coworkers (112) reported that in a dairy barn in whi(DDT has been applied as a space spray since 1945, resistant flies have not developed. In contrast, a hog barn about 0.8 mile from the dairy barn has been repeatedly sprayed with DDT for residual treatment and here a decided resistance has developed. Pimentel and Dewey (130) found that the larvae of a field-collected resistant strain were also resistant.

Barber and Schmitt (13) and Hansens and Goddin (63) found that in two colonies from the strain of flies used as a standard by the members of the Chemical Specialties Manufacturers Association (then the National Association of Insecticide and Disinfectant Manufacturers) there appear(to be resistance to methoxychlor. Such resistance has been reported by none of the many other laboratories using this strain.

The list of insecticides used in the laboratory to develop resistar strains of flies includes DDT (94), chlordane (99), gamma benzene hexachloride (23, 131), parathion (103), pyrethrum, toxaphene, methoxychlor, paraoxon, and dieldrin (33). Early in 1946, long before resistant flies were reported from the field, Lindquist and Wilson (94) began the development of the Orlando resistant strain. Flies from the regular laboratory colony were exposed to such a dose of DDT that about 90 per(

were killed. The survivors were used to carry on the colony. After three generations of such exposure, resistance was found. Essentially their technique has been used in the development of all the other laboratory-resistant strains reported in the literature.

In all but two cases noted, attempts to develop resistance in house flies by exposure to a given insecticide have been successful. The exceptions are the failure by D'Alessandro et al. (42) and the failure reported by Harrison (65) of attempts to develop a strain resistant to gamma benzene hexachloride from an Italian strain of resistant flies. In Harrison's case, only five generations were selected and this small number probably accounts for her failure. The development of strains resistant to benzene hexachloride, in the laboratory as well as in the field, is well known.

Attempts by Bucher, Cameron, and Wilkes (34) to develop a strain of house flies resistant to cold were unsuccessful.

Loss of resistance

Barber and Schmitt (14) tested the Ellenville line of resistant flies after 10 and 11 generations in the laboratory without exposure to DDT. These flies were still resistant to DDT, but not to the same degree, as the third generation. After 2 hours' exposure to 144 mg. of DDT per square foot, the 24-hour mortality reported for the third generation was 10.9 percent and for the "10th and 11th" (which one was not specified) 59.2 percent. March and Metcalf (100) reared the Bellflower resistant strain for 35 generations without exposure to DDT and without loss of resistance. They state that "At present no indication has been found that resistant strains will revert to more susceptible strains following non-exposure to the insecticide." More recent tests, however, (102) "have shown a trend toward an increase in the number of more susceptible flies in comparison with the number of highly resistant individuals." In the writers' laboratory a colony started from pupae furnished by Metcalf and March in November 1949 showed considerable resistance to insecticides in the first two generations. Within 10 generations without exposure to insecticide, however, the resistance had fallen to the level of that of the normal laboratory strain (unpublished results). At the Orlando, Fla., laboratory of this Bureau tests on the Bellflower strain showed that after 15 generations without exposure to insecticide a marked loss of resistance had occurred. The ratio, however, was still 10 times that of normal flies compared with 80 times when the strain was received (personal communication from W. V. King). The Orlando resistant strain loses resistance slowly but steadily if exposure to DDT ceases (72). Keiding and Van Deurs (71) found that the resistance of their strain decreased considerably after 7 to 9 generations although it was still remarkably high.

Knipling (77), speaking of work done at the Orlando laboratory, stated that 6 colonies of resistant flies collected in nature showed 20 to 50 times the resistance of normal flies to DDT. After 6 genera-

tions without exposure to DDT, some loss of resistance was apparent but the extent of loss was extremely variable and most of the flies still showed some resistance. King (72) reports that nearly all wild resistant stocks tested showed some loss of resistance when exposure to DDT was stopped. The rate of loss of resistance was very inconsistent. Bruce (30) on the other hand states "The several DDT-resistant strains of flies have shown no tendency to revert or lose their DDT tolerance through 34 generations of inbreeding." Bruce and Decker (33) state "All strains of DDT-resistant flies studied have retained their respective levels of tolerance when placed in a toxicant-free environment." They further state "It would appear by inference that if the chemical was entirely removed in the field, then the resistant strains might be effectively diluted by interbreeding with susceptible wild strains. This, of course, would be possible only if done before the entire population acquired a high degree of tolerance. Data obtained in a field survey may indicate this opportunity is rapidly passing if not gone." According to a later report, however, Bruce (6) now has experimental evidence that "dilution of one resistant strain with susceptible flies does not result in a strain with reduced resistant qualities." If this report is substantiated, the genetics involved are indeed unusual.

Specificity of resistance

Wilson and Gahan (170) tested the resistance of the Orlando resistant strain to several insecticides other than DDT, which was the insecticide used in the selection of the strain. The strain was more resistant to all the materials tested than was the regular colony. The materials were DDT, chlordane, pyrethrins plus 5 percent of piperonyl butoxide (for composition see 162), chlorinated camphene (toxaphene), rotenone, and thanite. Miller (109) compared the resistance to pyrethrins and pyrethrin mixtures of several strains of flies whose resistance had been developed by exposure to insecticides other than pyrethrins. No resistance to pyrethrins was found.

By exposing DDT-resistant flies (Orlando-Beltsville resistant strain) to a mixture of DDT, chlordane, toxaphene, lindane, methoxychlor, and pyrethrins, Pratt and Babers (134) developed a strain of flies that was resistant to these insecticides and to some extent to parathion as well. However, the resistance of this strain to the above insecticides was not statistically different from that of the Orlando-Beltsville resistant strain, whose resistance had been developed by exposure only to DDT.

Bruce (29) reported that strains can be developed to be more or less specifically resistant to one chemical or to closely related chemicals, but presented no data from which the degree of specificity could be determined. He found his resistant flies had a high tolerance for DDT and Rhothane D-3 (1,1-bis(p-chlorophenyl)-2,2-dichloroethane). A small amount of tolerance was evident for methoxychlor, and no "practical amount" for chlordane, heptachlor, toxaphene, aldrin, dieldrin, pyrenone, (piperonyl butoxide-pyrethrins mixture), or benzene hexachloride. He gave no data supporting these results. In his next paper Bruce (30)

presents data which indicate that the LD-50 of the most resistant strain is 776 times that of the normal strain for DDT, 14.4 times for methoxychlor, 3.2 times for lindane, 1.5 times for chlordane, 2.1 times for dieldrin, 1.6 times for piperonyl butoxide plus pyrethrins, 2.5 times for toxaphene, 9.9 times for DDT plus methoxychlor, and 1.5 times for paraoxon (diethyl p-nitrophenyl phosphate). Here too then, some resistance for nonrelated insecticides seems apparent. In a third paper Bruce and Decker (33) state "The high resistance of the DDT strain I to DDT ..... contributes resistance to other insecticides. DDT strains II and III with lower DDT resistance, show but little resistance to other insecticides." Earlier in the same paper, they state that DDT strain II was as resistant as DDT strain I. They state further "In general, the DDT-resistant strains exhibit a significant amount of tolerance for methoxychlor. On the other hand, the highly resistant methoxychlor strain shows little tolerance for DDT." Barber and Schmitt (13) also found no resistance to DDT in a strain of flies resistant to methoxychlor. Bruce and Decker continue, "The lindane-resistant flies show some tolerance for chlordane, dieldrin, and toxaphene but no significant resistance to DDT. This may indicate some similarity in the physical or chemical structure in lindane, chlordane, dieldrin, and toxaphene molecules." The chemical structure and physical properties of the above compounds show no readily recognized similarities except for the presence of chlorine. The basis for this suggestion then is not clear to the writers. Sufficient research has not yet been done to determine whether or not these compounds show any similarity of physiological action.

D'Alessandro et al. (42) suggest that resistance to "other insecticides" may be due to inability to penetrate the chitinous covering rather than a resistance to the insecticide itself." They intimate that there is possibly a difference in reaction between mists and residues. The reason for this suggestion is not clear, since the senior author and Scerrino (43) had shown that resistant flies are resistant to both sprays and deposits. Numerous other authors have, of course, shown that resistance is independent of the method of application. The authors at that time did not have sufficient data to state whether or not their super-resistant strain was also resistant to chlordane.

According to Brown and Rogers (28) the Ellenville DDT-resistant strain was markedly resistant to 1,1-dianisylneopentane, a compound in which three methyl groups are substituted for the three chlorine atoms in methoxychlor. Pimentel and Dewey (130) collected a field strain that was seven times as resistant as their normal strain on an LD-50 basis to DDT. There was possibly a slight resistance to methoxychlor, but none to dieldrin, gamma benzene hexachloride, aldrin, chlordane, or emulsifiable pyrenone (piperonyl butoxide-pyrethrins mixture).

The strain of wild flies collected in Denmark by Keiding and Van Duers (71) was resistant to DDT and its analogs. No resistance to benzene hexachloride, chlordane, or toxaphene was evident. King and Gahan (74) reported that the LT-70 (time of exposure in minutes required for 70 percent mortality) for females of a wild strain (Lee) was 11 times that of females of the normal strain for DDT, 2.6 for methoxychlor, 1 for chlordane, and 0.6 for benzene hexachloride.

March and Metcalf (99) state "The resistance of the Bellflower strain to gamma benzene hexachloride, toxaphene, chlordan-type compounds, parathion, and pyrethrins is small or inappreciable." Later (102) they find that "There are now two types of resistant fly strains in the field in southern California. The Bellflower type is resistant only to DDT and related compounds, and the Pollard type is resistant to other chlorinated hydrocarbons such as lindane and dieldrin as well as to DDT and related compounds." The data presented, however, indicate that for the Bellflower strain for which specific resistance to DDT and its analogs is claimed, the LD-50 is 8 times that of the normal laboratory flies for benzene hexachloride, 2.8 times for toxaphene, 1.9 times for heptachlor, 2.2 times for aldrin, 1.6 times for dieldrin, 1.3 times for parathion, and 2.3 times for allethrin. Dr. Metcalf, in a personal communication, explains that for "practical" purposes, they have not considered any insects as resistant until such resistance was 10 times that of the normal insects. These authors, therefore, evaluate resistance in terms of effectiveness of control rather than as a physiological difference between strains.

Knipling (78) reviewed some of the work that has been done on flies resistant to insecticides. He stated that tests have been made at Orlando to determine whether DDT-resistant flies were resistant to other insecticides, and the tests showed the flies had considerable resistance to methoxychlor, some slight resistance to chlordane, but little if any to benzene hexachloride. Apparently he referred only to wild resistant strains, since Wilson and Gahan (170), as indicated above, considered the "Orlando resistant strain" resistant to all other insecticides tested.

Pimentel et al. (130) state "When DDT-resistant flies were subjected to lindane, dieldrin, or parathion pressure and not DDT, the DDT resistance did not decrease, and in some cases even increased. If this occurs in the field, then the control of flies with residual materials is not a bright prospect." That such events do occur in the field is indicated by the work reviewed by Knipling (79), who, again referring to tests made at Orlando on a strain of flies collected from a local dairy, stated that the approximate magnitude of resistance compared to normal flies was as follows (assuming a level of 1 for the normal strain): "DDT 8, methoxychlor 4, chlordane 4, toxaphene 8, lindane 8, dieldrin greater than 16, and aldrin greater than 16. The flies exhibited no resistance to parathion, pyrethrins plus piperonyl butoxide, and allethrin plus piperonyl butoxide." The dairy from which these flies were

collected had been treated with DDT from 1945 through 1947. In 1948 one application of methoxychlor was made. In the spring of 1949 two treatments of pyrethrins plus piperonyl butoxide were made at 1-week intervals. Dieldrin emulsion was then used during the remainder of the 1949 season. In 1950 fixed aerosol equipment was installed for the daily application of lindane aerosols. A pyrethrum-piperonyl butoxide aerosol was then substituted, but results were unsatisfactory as most of the flies left the building before knock-down occurred.

Physiological and morphological differences between resistant and normal strains

Wiesmann (168) reported that the "Arnas" strain of resistant house flies had thicker pulvilli and articular membranes of the joints than did his normal laboratory strain, thus indicating that resistance was due to failure of a lethal quantity of DDT to reach a site in which physiological action would occur. Bettini (18) and March and Metcalf (99) agreed that when DDT solutions were injected directly into the hemocoele of the flies, the same relative degree of resistance was noted, thus demonstrating that resistance was not due to lack of penetration alone.

Hookum was quoted (7) as being unable to detect any structural differences between normal and resistant flies. D'Alessandro and his coworkers (42) measured the diameter of the pulvilli of the pretarsi of normal and resistant flies and found that on the average, the diameter of the resistant insect's pulvilli was less than that of the susceptible flies. It was clear, however, that the great difference in susceptibility was not proportional to the size of the pulvilli. March and Lewallen (98) compared normal and resistant (Bellflower) strains and found no gross morphological differences. In general the tarsi of the Bellflower strain tended to be shorter than those of the normal flies. The tarsal widths were variable. No significant differences in the thickness of the cuticula from the first tarsal segment were found. Neither was the slight difference in life cycle noted significant. The average life cycle of the Bellflower strain was 18.1 days and that of the laboratory strain 17.9 days. Over a period of 7 months, no difference in the weight of female flies was observed. Both strains were similarly immobilized by heat and cold, an indication that there was no significant difference in the general vigor of the two strains. The authors concluded then that neither thickness of the cuticula nor general vigor are concerned in the increased resistance of the Bellflower strain. "The differences in the dimensions of the tarsal segments of the two strains are not uniform enough or large enough to indicate they contribute to the resistance."

Neri (123) observed that when DDT was applied to different portions of the body of the fly, the resulting mortality varied greatly. However, when resistant flies were used, except for applications to the antennae, no differences were noted. Applications of DDT to the antennae of resistant insects were in no cases lethal. Wilson (169) also found

variation in the mortalities obtained from applying pyrethrum solutions to varying parts of the bodies of normal flies but did not use resistant insects.

Bruce (29) reported that the total development time from egg to adult of his resistant strains appears to be 1 to 2 days longer than that of the normal laboratory strain. He concludes that under field conditions the resistant flies would then tend to disappear when in competition with flies of a shorter life cycle. Bruce did not elaborate on the reasoning by which this conclusion was reached.

Pimentel et al. (131) also report that the life cycle of several strains of resistant flies was longer than that of non-resistant strains. The difference was most noticeable during the larval period and the length of this period seemed to be definitely related to the degree of resistance. "DDT-resistant flies from larvae which pupated the first 48 hours were less resistant to DDT than flies from larvae which pupated the last 48 hours in a given larval population for the strains studied. No significant differences between a highly DDT-resistant strain and a non-resistant strain were found in the number of eggs laid, length of egg stages, percent hatching, length of pupal stage, pupal weights, sex ratio, preoviposition period or length of adult stage."

Babers and Pratt (10) found that the cholinesterase activity of the heads of resistant (Orlando-Beltsville) adult flies was consistently lower than that of the normal flies for the first 5 days after emergence. The enzymatic activity of both strains with respect to acetyl choline was inhibited by excess substrate. Differences due to strain were not determined since only 1 resistant and 1 normal strain were studied.

Sacktor (144) determined the cytochrome oxidase activity of a strain of resistant flies derived from the Ellenville strain of Barber and Schmitt (14) and the normal laboratory strain of the Army Chemical Center. The enzyme activity of the brei from whole flies increased rapidly immediately after emergence. A peak was usually reached about the second day and a gradual decrease followed until about the fifth day. The changes after the first day were much more apparent in the resistant strain. The resistant strain consistently had a greater cytochrome oxidase activity than did the normal insects. In the normal strain, the male and female insects on a per fly basis had similar enzyme activities. However, on a weight basis, the male normal flies showed more oxidase activity than did the females. In the resistant strain, the females had about 20 percent more activity than did the males.

Sternberg, Kearns, and Bruce (154) applied ethanol solutions of DDT externally to the pronotum of both normal and resistant flies (Bruce and Decker (33) multi-strain I) and after a specified time the remaining external DDT was removed by washing. The flies were then crushed, extracted with ether, and the ether analyzed for DDT and the suspected metabolic products DDE (2,2-bis-(p-chlorophenyl)-1,1 dichloroethylene) and DDA (2,2-bis(p-chlorophenyl) acetic acid). The rate of absorption of DDT

was highest the first hour after application, possibly due to the effects of the alcohol used as solvent. The normal, or susceptible, strain absorbed DDT at a fairly constant rate for 4 hours. The rate then decreased sharply, coinciding with the appearance of paralysis. Unlike the susceptible flies, resistant flies continued to absorb DDT at a steadily diminishing rate so that by 28 hours after treatment, no DDT could be recovered from outside the body when the original dosage was 0.5 microgram per fly.

In the susceptible flies, analysis showed that there was a very slow metabolism of DDT but that neither DDE nor DDA were metabolic products. However, in the resistant strain, the absorbed DDT was rapidly metabolized, essentially to DDE but some DDA was also found. The evidence for the identification of the metabolic products was based mainly on spectrophotometric data and cannot be considered as conclusive. After 54 hours a group of 20 flies that had metabolized over 50 micrograms of DDT had only excreted 5 micrograms of DDE. The method of metabolism was not determined. In vitro tests for metabolism were inconclusive. It seemed possible that the resistant flies were able to metabolize DDT before it reached its site of physiological action.

More recently Sternberg and Kearns (153) have expanded their work on the metabolism of DDT to a considerable degree. In an attempt to determine the site of degradation of the DDT, tissues from several portions of house fly anatomy were analyzed. After the external application of DDT to resistant flies, the degradation product DDE was found in most areas of the body but was most abundant in cuticle from the regions of the body to which the dosage was applied, thus suggesting the possibility that the DDT was degraded to DDE during the time it was being absorbed through the cuticle-hypoderm.

If the mouth parts, legs, and wings of resistant flies were removed before the DDT was applied, neither DDT nor DDE was found in any of the internal tissues of the flies. DDE was found only in the entire head and thoracic and abdominal cuticle. Further experiments showed that if DDT was fed the flies in milk (2 mg. of DDT per milliliter), it was degraded in the digestive tract. The DDE produced was transported to all parts of the body, but non-metabolized DDT was retained in the digestive tract and did not accumulate in other parts of the body. The ability of resistant flies to degrade oral doses of DDT before any has reached a vital site thus appears to be a further factor in resistance of DDT. Whether or not susceptible adult flies were able to degrade DDT in their digestive tract was not determined. In view of the work with larvae mentioned later, this question is of considerable interest.

If DDT was applied to susceptible flies following removal of the mouth parts, only 70 percent of the 0.5 microgram dose applied was recovered; 42 percent of the recovered DDT was found in the thoracic cuticle, legs, and wings, and 28 percent was still on the external portions of the fly. Neither DDT nor any metabolite that responds to the

Schechter-Haller test was recoverable from the internal tissue of the fly. The authors were unable to confirm the results of Läuger et al. (84) and Bot (25), who, using bioassay methods, recovered a material toxic to other flies from the thoracic ganglia, Malpighian tubules, and gut of DDT-poisoned flies. The material was presumed to be DDT. Lindquist et al. (93), as reported below, found that both susceptible and resistant flies rapidly metabolized DDT to a product nontoxic to mosquito larvae. Sternberg and Kearns (153) found that both susceptible and resistant flies absorbed DDE at comparable rates and neither strain was able to further metabolize this compound. Both strains were also unable to further metabolize DDA. Degradation of DDT to DDE in vitro by the cuticle-hypoderm of resistant flies was demonstrated, but the conversion was below 20 percent. No other tissues tested were able to degrade the DDT. It was also found that resistant larvae could metabolize externally applied DDT to DDE and store it in considerable quantity. Susceptible larvae on the other hand metabolized a considerable portion of the DDT but the metabolite, as in the case of the adult, was unknown. It was not, however, DDE or DDA. When 10 micrograms of DDT were applied to each of 10 susceptible and 10 resistant larvae, the data presented indicate that the susceptible larvae absorbed the DDT at a very much greater rate than did the resistant larvae. The authors did not comment on this difference. When larvae were placed in medium containing DDT for varying periods and then analyzed for DDT and DDE, it was found that both DDE and DDT were present on the external portions of the resistant larvae. Only DDE was found internally in these larvae. On the external portion of susceptible larvae, only DDT was found, but internally both DDT and DDE were found. If the DDT-fed larvae were allowed to pupate and the pupae analyzed, similar results were obtained. The resistant adults emerged normally and contained DDE both externally and internally. The few susceptible adults that emerged contained DDE internally only. If DDT was applied topically to resistant pupae, they also were able to absorb and degrade it to DDE. Susceptible pupae were also able to degrade DDT to DDE to some extent.

In many of the above experiments, the quantities of DDT and DDE determined were considerably below those for which the analytical methods available are usually considered accurate. The authors, however, apparently encountered no difficulty in this respect. Perry and Hoskins (128) found that DDT absorbed by both normal and resistant flies was metabolized to what was first thought to be DDA (5) but later presumed to be DDE (128). This was contradictory to the results of Sternberg et al. (154) who found that only a resistant strain could metabolize DDT to DDE but agreed in general with the results reported by March and Metcalf (103). From the normal strain in one experiment Perry and Hoskins recovered 30 percent of the amount of DDT applied externally as internal DDT and 66 percent as DDE. With the resistant strain the applied DDT recovered as DDT was 7.5 percent and DDE 33.1 percent. These authors conclude "It is obvious that increased ability to convert absorbed DDT into DDE is characteristic of the resistant strain." The authors analyzed both survivors and dead insects and found "In a given experiment, the survivors on the average always had converted more DDT

than those that had died, and hence ability to make this conversion is a major factor in variation in resistance within individuals of a given strain." From dead resistant insects treated with DDT plus piperonyl cyclonene, only 3.4 percent of the applied DDT was recovered as DDE contrasted with the 17.5 percent without the synergist. They conclude then that in flies treated with DDT-synergist mixture the conversion to DDE was largely prevented. They state also that the total recovered DDT from the resistant strains was at least a third less than that applied. March and Metcalf (102) tested the compounds commonly used as synergists for pyrethrum to determine the effect on DDT-resistant flies. Little or no effective synergism for DDT was observed. They (103) also studied the metabolism of DDT and found that the super-Bellflower strain of resistant flies was able to metabolize larger quantities of DDT at a greater rate than were normal flies. DDE was the principle degradation product and there was no evidence of the formation of DDA. The end products as yet have not been completely defined.

Lindquist and his associates (93), using radioactive DDT and bioassay methods, have found that DDT applied externally to flies is slowly absorbed. Once absorbed, however, it is rapidly metabolized by both normal and resistant (Orlando) insects to a material nontoxic to mosquito larvae.

In the writers' laboratory (unpublished results) little difference in mortality was obtained when dosages varying from 50 micrograms of DDT per fly to 300 micrograms were applied externally to female resistant (Orlando-Beltsville) flies. The volume of solution applied was 1.14 microliters per fly. From the first dosage the mortality at 24 hours was 18 percent and from the highest dose 16 percent. Only a small percentage of the DDT absorbed was accounted for by analysis of extracts of the acetone-washed flies. From such extracts small amounts of DDT and a compound presumed to be DDE were recovered. In no case was DDA found. Results obtained from the analysis of normal flies have been inconclusive to date. In some cases small amounts of DDT and DDE have been recovered and in other cases neither material was found. The Schechter-Haller method of analysis which was used is not well adapted to the small quantities of DDT involved when normal flies are used, and some difficulty with interfering materials was encountered.

Transmission of resistance

Bruce (29) states "Further study with the resistant flies has shown by several reciprocal crosses of susceptible and resistant flies that resistance is neither dominant nor recessive, but rather than being a gene characteristic is probably a cytoplasmic characteristic." No data was given to support this statement. Bruce (29) and Bruce and Decker (33) interpret data obtained by crossing resistant insects with normal insects to mean that both male and female flies carry the resistant character. "Tolerance might simply be described as a multiple-gene character which causes indifferent physiological and perhaps morphological changes." Later in the same paper they state "There is reason to believe

that resistance to toxicants by house flies may be attributed to indifferent genetical changes, ephemeral acquired tolerance, morphological alterations or even changes in habit." The genetical data presented is limited to the LD-50 of the male and female progeny of the F_1, F_2, and F_{15} generations resulting from the reciprocal crossing of a resistant and normal strain of flies. Bruce, in a discussion of Knipling's paper (78), commented that information on crossing strains shows not so much a Mendelian inheritance as a dilution or cytoplasmic inheritance. Bruce (29), Decker (6), and Bruce and Decker (33) observed that "fly resistance to DDT is not all chemical in nature" and that in certain insecticide-treated barns, house flies no longer rested on the treated surfaces but preferred to rest on the floor or on the animals. No data were presented.

D'Alessandro and his coworkers (42) started a strain of susceptible flies from a single pair of flies captured in a locality which had not been treated with DDT. This strain (partinicensis) showed a relatively homogeneous sensitivity to DDT and all individuals were killed after a 35-minute exposure to DDT-treated cages. In a strain (tiberina) secured from Missiroli's laboratory, the knock-down time varied between 20 minutes and 2 hours and thus contained both susceptible and resistant individuals. After repeated selection to eliminate the susceptible individuals, a strain (supertiberina) was obtained capable of living many days in DDT-coated cages. On the other hand, the attempt to select a resistant strain from partinicensis was unsuccessful. Apparently the strain had been deprived of resistant characters. The authors were unable to increase resistance in individual flies of the strain partinicensis by repeatedly exposing them to nonlethal doses of DDT. The toxicity of DDT was not accumulative after such series of exposure, an indication that the flies were able to detoxify or metabolize the material. The authors define as susceptible flies those unable to survive 35 minutes in a DDT-treated cage, as medium-resistant those able to survive 1 to 2 hours, and as super-resistant those able to survive 24 hours in the cage. In a study of the transmission of the resistance, sufficient data was not obtained to establish a strict definition of a phenomena within the laws of heredity. However, the strain of susceptible flies remained homogeneously susceptible. The strain with individuals of varying resistance so remained, and the super-resistant strain remained homogenously resistant. By pairing a super-resistant female and a sensitive male, 177 individuals were obtained, 5.5 percent sensitive, 90 percent medium-resistant, and 4.5 percent super-resistant. The mixed generation thus produced was allowed to interbreed promiscuously and the F_3 generation consisted of 24.2 percent susceptible, 73.1 percent medium-resistant, and 2.7 percent super-resistant. All the super-resistant flies were females. The arrival then of super-resistant female flies in a sensitive population would lead to a diffusion of the resistant character. The authors concluded that the phenomenon of DDT resistance does not represent a definite quantitative characteristic but one that is extremely varied in degree. Also that by a series of selective isolations beginning with moderately resistant individuals, it is possible to

obtain generations that are highly resistant to the poison. The data presented in the manuscript are very meager and few of the techniques used are described. According to the authors, supporting data will be published elsewhere. Until such information is available the work cannot be properly evaluated.

In the writers' laboratory, after about 92 generations of intensive selection by exposure of each generation of house flies to DDT sprays or residues, the population is still by no means homogeneous (unpublished results). Pupae to start this line of flies, now designated Orlando-Beltsville resistant, were from the 55th generation of the Orlando resistant colony. For about the last 20 generations selection has been made by holding adult flies in cages coated with continuous deposits of DDT (about 2 grams per square foot) for several days after their emergence. The colony is maintained from the survivors. Although the percentage of survivors has increased, many flies are still killed by the treatment.

Control of resistant flies

That the extent of development of resistance to insecticides by the house fly was not quickly realized by all entomologists is indicated by Hatfield's (66) statement in 1949: "Personally, I think DDT produced in 1948 or 1949 applied like the 1946 and 1947 DDT was applied, will give results comparable to the 1946 or 1947 results." He implied that if DDT was applied at the rate of 200 mg. of DDT per square foot it would give excellent control of resistant flies. The many laboratory reports of testing resistant strains against this and even higher dosages shows the error of his supposition.

Following the failure of DDT to control flies in certain areas, recommendations were often made to change insecticides. Ayars (8) stated "Fortunately flies resistant to DDT are relatively susceptible to lindane, chlordane, toxaphene, and dieldrin. ... Entomologists expect lindane to be effective against house flies for about 2 years. By about 1952 or 1953 perhaps a new insecticide will be needed." Mosna (115, 116, 117), Patrissi, Barbieri, and Bessler (127), Bettini and Barachini (20), and others (1, 2, 19, 44, 57, 141) agreed that the DDT-resistant flies could be effectively controlled with chlordane, lindane, or toxaphene. Decker (6) proposed alternate use of lindane, chlordan, and dieldrin. Instead of the 2 years' effectiveness predicted by Ayars for lindane, March and Metcalf (100, 101, 102) found that in the field several DDT-resistant strains had in a few months become resistant to lindane and dieldrin as well as to DDT. They state (101) "These new developments, though as yet only on a limited scale, indicate that eventually standard fly control procedures may have to be re-evaluated and revised, and that emphasis may have to be placed on sanitary measures, repellent materials, and space sprays, rather than on residual application of insecticides." Fjelddalen (52) discussed DDT resistance and the use of methoxychlor as a control chemical for the resistant flies.

Gahan and Weir (55) used gamma benzene hexachloride for the control of Musca domestica vicina Macq. in Egypt. It was not stated whether or not other insecticides had ever been used in the area. For the first 10 months, fresh applications were made once a month with very effective results. After 1 year, the intervals between treatments were shortened to 1 week and even then effective control was not obtained. Laboratory tests confirmed the field tests and showed that resistance had developed. No other insecticides were tested. The authors conclude that "the results obtained ... indicate that any benefit derived from a change to this insecticide (in order to control DDT-resistant flies) might be only temporary."

Bruce (30) stated that the substitution of methoxychlor for use against DDT-resistant flies gave unsatisfactory results due to the rapid acquisition of tolerance for the compound. Bruce (6) suggested that "extensive use of one insecticide will produce a tolerant strain more quickly," and later he (31) stated "Probably the development of insecticide tolerance can be avoided by not relying upon a single highly effective material for the control of any particular pest. Use a chemical to supplement other kinds of control methods and change chemicals as soon as there is any evidence of resistance developing to an insecticide." To date little evidence has appeared which indicates that changing residual type insecticides under natural conditions will in any way prevent the appearance of resistant strains.

Catalano and Mariani (39) report that the addition of fats or oils to DDT solvents remarkably increases the toxicity of the material and that "flies that are most resistant to residuums of the usual solutions of DDT behave like sensitive flies on being exposed to residuums obtained from solvents containing olio. Davidovici et al. (45) also report that the addition of lanoline to DDT-kerosene solutions enhances the toxicity of the residue to resistant flies and mosquitoes. The increased toxicity was lost after about 3 weeks when crystals of DDT began to appear in the residues. D'Alessandro et al. (42) report that 5 gamma of DDT in a fat solution is lethal to super-resistant flies that, according to Bettini (18), are able to survive 5 gamma in olive oil when injected directly into the thorax.

Perry and Hoskins (128) found that piperonyl cyclonene (for chemical composition see 162) increased the toxicity of DDT for two strains of resistant flies. Thus 5 micrograms of DDT alone per fly gave 42.1 percent mortality and 5 micrograms of DDT plus 25 micrograms of piperonyl cyclonene gave 87.9 percent. If the synergist was increased to 50 micrograms, the mortality dropped to 76.8 percent. With the normal strain, no synergism was observed. This work has not yet been confirmed.

Because DDT failed to control flies due to the development of resistance and partly because of recommendations that DDT be avoided in dairy barns or on dairy animals, Hansens (62) investigated several other insecticides. Lindane or methoxychlor was recommended as replacement for DDT in these circumstances.

According to Upholt (160), "the often expressed opinion that the situation could be solved by rotating insecticides is ... without foundation." He believes that resistant strains whose degree of resistance is low will lose resistance if kept out of contact with insecticides. Practically, "it is a vain hope to expect under field conditions to maintain any population of flies out of contact with DDT, or with any other insecticide, which is used to such an extent as to become a factor in resistance." A strain of flies from an area in which resistance was pronounced, quickly lost their resistance in the laboratory when not exposed to DDT. In the field, however, after more than a year after stoppage of the DDT program, the resistance was not diminished. The explanation of course was that the stoppage of the use of DDT for fly control did not stop the use of DDT in the area for other agricultural pests and did not remove the DDT from any areas in which large quantities had been applied. In Upholt's opinion, the development of resistant insects had been greatly accelerated by the substitution of DDT for adequate sanitation as a fly control measure.

Other Species

In Illinois, filter flies (*Psychoda alternata* Say) were no longer controlled by treatment of the larval habitat with DDT, according to Bruce (30) and Bruce and Decker (33), after 2 years of excellent control. The first chlordane treatment was highly successful, the second only moderately so, and the third and fourth were near failures. Schaerffenberg (145) studied the absorption of DDT and the resistance to the compound in certain May beetles.

Contrary to results in the United States, Raucourt (136) reported that lead arsenate was still very effective in France for the control of the codling moth. According to him, the failure of lead arsenate to control the insect in the United States had not, in published reports, been attributed to resistance. This statement is in error, since several publications have appeared, as noted in the earlier review (9), on the subject of such resistance. More recently Palm (126) states that the entire Hudson Valley orchard area has developed a codling moth problem that lead arsenate will not handle. He quotes S. W. Harman as finding in 1945 that only the very high prices received for the fruit at that time enabled growers in western New York to make a profit because of the failure of lead arsenate to control the insect. Up to 1949 it was the opinion of several entomologists familiar with the subject (3) that no case of codling moth resistance to DDT or of other DDT-resistant fruit insects had been proved. No more recent references have been noted.

Pielou (129) states that "Investigations are in progress to determine the possibility of breeding strains of beneficial insects resistant to DDT and other materials." These attempts have not progressed far enough to predict their outcome.

That resistance to lead arsenate by the peach twig borer (Anarsia lineatella Zell.) had developed in a small area in California was suspected by Bailey (11). Laboratory experiments by Summers (155) tended to confirm these suspicions. No tests have been conducted with other insecticides, nor is it clear whether economic control of the insect with lead arsenate is still indicated.

Shepard (146, p. 327) quotes G. Peters as finding that an exposure to a small quantity of hydrocyanic acid causes a stupefying effect on the granary weevil (Sitophilus granarius (L.)) that results in cessation of respiration. The cyanide can thus only enter the insect by diffusion and hence the high resistance of the species.

Kono (82) found that prefumigation of the rice weevil (Sitophilus oryza (L.)) with low doses of carbon disulfide for 24 hours lowers susceptibility. There was no significant difference in oxygen consumption. Berim and Edelman (16) studied the resistance of several insects to benzene hexachloride and DDT and found that it varied greatly with time of day and the season.

Ricci (138) studied the action of DDT on the oriental cockroach (Blatta orientalis L.) and found that strains obtained from areas previously treated with DDT showed increased resistance to the insecticide.

In a colony of body lice (Pediculus humanus corporis Deg.), after the exposure of 8 generations to DDT, no increased resistance to the insecticide was noted (72).

According to D'Alessandro et al. (42), Mariani found that after two years of treatment with DDT the dictyospermum scale (Chrysomphalus dictyospermi (Morg.)) had become resistant to a considerable degree. After the first treatment with DDT only 2 to 5 percent survived, but after 2 years of treatment 25 to 30 percent survived equivalent dosages.

Johnson and Hill (69) report that in Hawaii bed bugs (Cimex lectularius L.) have become resistant to DDT.

Stanley W. Bromley (personal communication), from the results of field tests, has concluded that the bark beetle (Scolytus multistriatus (Marsh.)), the carrier of Dutch elm disease, has developed a considerable degree of resistance to DDT and to a lesser extent to methoxychlor. The beetles were from an area that had been treated with DDT for several seasons.

According to Jayewickreme (68) field-collected larvae of Anopheles subpictus Grassi showed resistance in laboratory tests to films of mineral oil (Shell Malariol). The resistance was attributed to increased vigor caused by favorable environmental conditions.

A strain of the cockroach (Blattella germanica (L.)) resistant to DDT has been developed by J. M. Grayson (personal communication).

Following 7 generations of selective exposure to DDT, 14.3 times as much DDT was required for an LD-50 as was necessary in the unselected insects. Following 7 generations of selective exposure to benzene hexachloride, only a slight increase in tolerance was noted. In the DDT-resistant insects, no difference in reproductive performance was noted but the average number of nymphs per egg case was lower in the DDT-resistant strain than in the normal strain.

A species of bug, species not given but apparently the large milkweed bug, Oncopeltus fasciatus (Dall.), has been reported by Turner (159) as developing resistance to nicotine in the laboratory.

Discussion

Melander (108) is generally credited with the first publication on insects showing resistance to insecticides. According to Bruce (32), Piper in 1913 found the San Jose scale to be resistant to lime-sulfur. The reviewers have searched the usual reference sources but have been unable to locate a reference to this work by Piper. However, it appears that John B. Smith (152) observed resistance some years previously. Smith in 1887 stated "Insects which succumb readily to kerosene in the Atlantic States defy it absolutely in Colorado, while we are just as likely to find the food plant much more sensitive to it. Washes that easily destroy the San Jose scale in California are ridiculously ineffective in the Atlantic States. This very scale is changing its life history and habits in the East materially in several directions. I will venture the prediction that in half a dozen years it will not be considered a first class pest in New Jersey, though I would not like to extend this prophecy to localities with which I am less familiar."

Monro (111) has discussed the subject of resistant insects and speculates to some extent on various aspects of the problem.

Variations within insect species was suggested in 1864 by Walsh (163). Certain wood-boring and plant-feeding beetles could to some extent be divided into groups by food preference. They occurred in the same locality and had definite differences in biology, but often showed no or at the most very minor structural differences. Thorpe (158) and later Smith (151) reviewed the occurrence of biological races in insects. In some insect species, the European fruit lecanium (Lecanium corni Bouche), for example, the external appearance varies considerably depending on the host plant. On the elm this scale has a large pruinose form and on the arbutus it is small and shiny. Ebeling (49), although unsuccessful in the field, was able to transfer the insect from its normal host to other plants in the laboratory. Succeeding generations conformed to the appearance of those insects regularly found on the new host plant.

No report concerning the development of resistance to insecticides by the hessian fly (Phytophaga destructor (Say)) has been observed. Some of the work with this insect, however, seems to warrant comment in this review, because of the possible bearing on the resistance problem. Data presented by Painter (125) in 1930 tended to show that the hessian fly population in any one locality is a mixture of two or more genetically distinct strains that seem to differ only in their ability to infest different varieties of wheat. Cartwright and Noble (38) further explored

this observation and found that the Dawson variety of wheat was resistant to the hessian fly in California and Kansas but was susceptible to the fly in Indiana. Thus the California and Indiana strains of flies seem to be physiologically different races. No reference has been noted indicating the effect of testing the California strain on Dawson wheat in Indiana or that of the Indiana strain on Dawson wheat in California.

There have been a number of reports of insects, particularly house flies, having specific resistance to one insecticide or its analogs. Other reports indicate that high resistance to one insecticide confers some resistance to most other insecticides, regardless of structure. The two apparently divergent views would be reconciled to a considerable degree if a standard definition of the term resistance and uniform methods of testing were adopted. Quayle (135) proposed that an insect species be termed resistant if there was a decreased effectiveness of an insecticide compared with that of former years. This definition seems to the writers to infer at least a direct relationship of resistance to economic control. Tests under field conditions have many variables that determine to a considerable degree the effectiveness of an insecticide. Under laboratory conditions several of the more important variables are eliminated, temperature, humidity and nutrition of the insects being relatively constant. The writers have considered that two strains of insects vary in resistance if, when tested under similar conditions, there is a statistically significant difference in their response to a given quantity of insecticide. Physiologically, a slight variation in response may be of considerable importance but at the same time have no appreciable bearing on the effectiveness of the material as an economic poison, since complete control may still be obtained with readily usable quantities.

In some cases the dosage of insecticide used has been such that all insects of the strains tested were killed and the conclusion reached that there was no difference in resistance between the two strains. It is obvious that such data cannot be considered a valid test for resistance.

Small differences in rearing methods might often account for some differences in results between laboratories. Swingle (156), Markos and Campbell (104), and McGovran and Gersdorff (107) have pointed out the effect of variations in susceptibility resulting in changes of diet. It is also necessary of course that temperatures to which the insects are exposed during the tests be relatively constant and also comparable to those used in other laboratories. Lindquist and his associates (92, 95) have shown the great effect of temperature on the knock-down and mortality obtained from given doses of DDT.

Many factors other than resistance often cause the failure of an insecticide to effect control. The term resistance then should not indicate in any way failure to obtain economic control. When house fly

resistance to DDT was first observed, the failure to recognize as danger signals the small increases in resistance to other insecticides led to many such statements as "DDT-resistant flies are readily controlled by", etc. Such statements, while perfectly correct at the time, in only a few instances carried warnings that such control would probably be only temporary. Indeed, if the data of Wilson and Gahan (170) showing that the Orlando resistant strain was also resistant to several other unrelated insecticides had not been largely ignored, researchers concerned with the field control of flies might have recognized that substitution of other highly toxic residual insecticides would result only in short term effectiveness.

At present, however, it seems that almost any positive statement concerning resistance will probably have to be rescinded or modified. The thought has been expressed in several publications that resistance follows repeated exposure to insecticides. Resistance of the red scale to hydrocyanic acid appeared in several isolated areas in California in 1916. Effective control of these resistant insects was obtained with oil sprays and the use of hydrocyanic acid was gradually supplanted by them. One would have expected that the resistance to hydrocyanic acid would disappear after the continued use of oil. However, the scales in these areas today are still resistant to hydrocyanic acid and the areas are appreciably larger than they were in 1916. Oil sprays are still used on them, and no resistance to the oil has apparently developed. Also after 25 to 30 years use of oil sprays against the San Jose scale, there is no evidence that lime-sulfur resistant scales have developed resistance to oil sprays. However, as Upholt (160) points out, the scale insect as well as the codling moth are highly specialized species restricted to a limited number of host plants. Especially with the scale insect, the species is not particularly adaptable. On the other hand, the house fly has adapted itself to all conditions to which man has adapted himself. Thus the house fly might be expected to develop resistance, whereas with the scale insects the development of resistance to hydrocyanic acid might be considered an accident of nature. Such a specialized insect would not be expected to rapidly adapt itself to changes in environment. "The house fly has a very high biotic potential, and when a population seems essentially to be wiped out, a very few survivors are able to restore the population rapidly to its normal size." "Adaptability is associated with a high frequency of gene mutation: and when these adaptations do become inherited through mutations, we have a situation which may be similar to the observed resistance in flies." Citrus thrips became highly resistant to tartar emetic in a comparatively short time but have not become resistant to other insecticides used before or since the introduction of tartar emetic.

Another anomaly is the arsenic-resistant tick. Apparently only one species is so far involved. Yet the infested cattle have been heavily infested with several other species of ticks that, although treated in the same manner, are still readily susceptible to arsenic.

Why, too, should the resistance largely disappear when the tick-infested cattle were moved inland? Similar anomalies occur in the case of the house fly. Strains that maintained resistance on the West coast for many generations lost the resistance rather rapidly when pupae were sent East to start a colony. Other cases have occurred in which apparently contradictory results were obtained when pupae or eggs from resistant strains of house flies were sent from one laboratory to another. Errors in sampling and even misidentification of strains have been suspected. To the writers, it seems more logical to assume that the results as obtained are correct and that more attention should be given to the indications and interpretations of such data.

The great difference in response to insecticides by the various populations of flies is emphasized by Upholt (160). He reports that in two areas in which dieldrin applications were made late in 1949 or early 1950, a high degree of resistance was soon developed and essentially eliminated the practicability of residual chemical control of flies in those areas with known insecticides. However, there are areas other than the above two in which dieldrin has been used for a longer period and with more frequent applications but no resistance is apparent. All populations therefore do not develop resistance to dieldrin at the same rate, but it seems probable that within a year from the time of its first use resistance can be expected.

At times positive statements have been widely disseminated that, from the results published in the literature, seem to have little or no data to support them. In a recent editorial (4) it is stated "Now the latest reports hint that after 20 or so generations of immunity (to insecticides) by flies, DDT and other chlorinated insecticides again become effective." There has been no evidence published that in the slightest degree seems to indicate such a trend. It may be that the author intended to say "20 or so generations without exposure to insecticides" but that is not what was published.

Plackett and Hewlett (132) and Hewlett and Plackett (67) statistically treated data obtained following the application of mixtures of two insecticides to insects, and discussed the effects on resistant insects of such mixtures if the two toxicants acted independently. According to their reasoning, if two insecticides having independent physiological action were applied jointly to insect populations, the development of strains of insects highly resistant to either insecticide would be prevented because the insects most resistant to each toxicant would be killed: only those insects with intermediate resistance would survive and any survivors could be expected to give rise to descendants also of intermediate resistance. No error in the statistical treatment is at once apparent, but it is difficult to understand how a dose of insecticide which kills the tougher individuals of a population would fail to kill the weaker ones first.

For many years insecticides have for convenience been classified according to their generally accepted mode of entry into the insect as

stomach poisons, contact poisons, or fumigants. The implication has sometimes been made erroneously that the mode of entry really constitutes mode of action. There seems to be widespread misconception that changing the mode of entry of the insecticide into resistant flies would drastically change or even overcome resistance. Since it has been shown that resistance is maintained when the insecticide is injected, physical factors such as thickness of integument or lack of penetration are not the cause of resistance in house flies and once an insecticide reaches its site of action, no evidence has ever been presented to indicate that mode of action varies with mode of entry.

The role that the mixing of various strains of insects in the field plays in the development of resistance has yet to be determined. However, because Bishopp and Laake (21) reported that marked house flies were recovered as far as 13 to 14 miles from the point of release, considerable effect must be expected. In other species of insects even greater dispersions are found.

No adequate genetical study of resistance in the house fly has yet been reported in the literature. Bruce (29), Bruce in Knipling (78), and Bruce and Decker (33) presented a hypothesis for the method of transmission of inheritance. To date, however, their published data consist only in the determination of dosage-mortalities for the F_1, F_2, and F_{15} generations resulting from the reciprocal crosses of a strain of normal with a resistant strain of flies. Whether such crosses were made with individuals or whether they were mass crossings is not clear. Neither is it clear how the meager data presented justify the conclusions made.

According to D'Alessandro and his associates (42) DDT sensitivity and DDT resistance are hereditarily transmitted to the progeny and apparent modifications are not met with in the course of many generations. Here again, the data so far presented do not seem sufficient to justify the drawing of definite conclusions.

The difficulties involved in the interpretation of data obtained from mass crossings are considerable. Several questions that must be considered in mass crossing are: What percentage of the population is resistant? What is the percentage of susceptible individuals? These questions apply equally to the normal strain, for it is well known that all insects of a given population do not respond in the same way to the same dose of insecticide. Are the results obtained from mass crossings with subsequent determination of the LD-50 of the F_1 population comparable to results from individual crossings with determination of the resistance of each individual progeny? How can the resistance of an individual insect be determined?

It has been generally accepted that antigens, the substances that disturb protein synthesis in the organism in such a way that antibodies are produced, are macromolecules and are generally proteins or protein

complexes. However, Loiseleur (96, 97) has reported that antibodies are formed in the serum against such simple substances as ethyl alcohol, ethylamine, sodium acetate, xylose, and arginine. The presence of the antibodies was demonstrated, except in the case of sodium acetate and ethylamine, by an increased viscosity of the serum after addition of the injected substance. When sodium acetate and ethylamine were used, flocculation was observed. Viscosity tests are generally not accepted as conclusive proof of the formation of antibodies. In the event that Loiseleur is correct in the interpreation of his results, his findings may elucidate to a considerable degree the phenomena of the development of resistance to various materials by vertebrates and possibly invertebrates as well. Such an explanation might also explain certain cases of cross tolerance, since the injection of a uniform antigen solution leads to the formation of multiple antibodies directed against the different determinant groups of the antigen molecule.

In the recent literature there are numerous references to "DDT and other chlorinated hydrocarbon insecticides." Often it is obvious that the author considers that "chlorinated hydrocarbon insecticides" are closely related chemically and should be expected to behave in a similar manner physiologically. Possibly this reasoning is based on the assumption of the correctness of the hypothesis of Martin and Wain (105) concerning the mode of action of DDT. According to this hypothesis, the chlorophenyl rings confer lipoid solubility on the molecule whereas the remainder of the molecule liberates hydrochloric acid at vital centers and is responsible for toxicity. Although such a possibility must be considered, little experimental data have appeared in its support and much of the data suggest that the theory is not correct. Dieldrin and aldrin, for example, are excellent chlorinated hydrocarbon insecticides but are little affected by strong alkali *in vitro*. Such stability *in vitro* does not preclude breakdown *in vivo* but the probability is strong.

Admitting for a moment the possibility that the toxicity of DDT was due to the liberation of hydrochloric acid, it would seem probable that DDT-resistant flies would be generally better buffered than the susceptible insects. The authors and M. Beroza of the Division of Insecticide Investigations (unpublished results) therefore ran titration curves on the brei obtained by grinding whole flies in distilled water under deobase. No difference was noted between the curves obtained from the brei of normal (Beltsville normal) and resistant flies (Orlando-Beltsville). Further doubt on the validity of the theory arises when one considers the isomers of DDT.

It is well known that a shift of the chlorine atom on one of the benzene rings of the DDT molecule to any other position markedly lowers the insecticidal value of the compound but these isomers readily dehydrochlorinate. Several hundred analogs of DDT have been prepared and in none of them has the insecticidal value of DDT been approached. Methoxychlor and the fluorine analog of DDT are also often included in lists of such "chlorinated hydrocarbons." To assume that all chlorinated hydrocarbons should act pysiologically in a similar manner is

about as logical as assuming that all plant material should act on insects in a manner similar to pyrethrum. The situation is further confused by the several reports that methoxyclor-resistant flies are not resistant to DDT, but that DDT-resistant flies usually show high cross tolerance for the methoxy compound.

Haarer (60), speaking of the development of resistance to benzene hexachloride by the already arsenic-resistant blue tick, states: "It seemed that news such as this quietly trickling through scientific circles, and appearing in small sections of the overseas press, is enough to make headlines as big as those concerning the new atomic bomb, if only the significance of the matter were properly understood."

In the United States flies, mosquitoes, cockroaches, and bark beetles have been definitely reported as developing resistance to the organic insecticides currently used in large quantities for the control of insects by contact with residual deposits. The development of resistance to benzene hexachloride by the blue tick in South Africa, of Drosophila to DFDT, and of bed bugs to DDT, proves that other species can and have developed resistance, and indicates further that the development of similar resistance by more species can be expected to occur in nature at any time.

It should also be pointed out that all work so far reported on the development of resistance in insects has been done on populations; never, apparently, has resistance been developed in an individual insect.

In conclusion it may be pointed out that the phenomenon of resistance is not much better understood now than it was at the time of the first report of its occurrence. The method of development or of transmission is not clear, and no adequate suggestion has been made as to what should be done to overcome the resistance. An extensive and coordinated long-range program seems to be indicated, but currently most of the research efforts seem to be directed toward temporary expedients rather than toward the solution of the problem. If one thing in the picture is clear, it may be summed up in Decker's suggestion (46)--"Remember what has happened to DDT and proceed cautiously in the use of other insecticides."

It is obvious from the various reports reviewed that there is variation between the response of the various strains, both normal and resistant, to the several insecticides. It is not possible, apparently to make any statement that will cover all cases but the following generalities begin to appear:

1. A very high level of resistance to one insecticide confers a certain amount of cross tolerance for other insecticides of widely dissimilar structure.

2. When the level of resistance is low, the resistance is almost

specific and is limited to other insecticides of closely related structure.

3. Strains of house flies resistant to all insecticides for which the attempt was made have been developed in the laboratory. This includes materials of plant origin as well as synthetic organic materials.

4. The resistance level of wild strains of house flies is by no means as high as that of laboratory-developed strains.

5. The degree of resistance in house flies varies considerably from generation to generation.

6. Most strains of resistant house flies, either laboratory or field, tend to lose their resistance if exposure to the chemical ceases, but some strains maintain resistance for many generations.

7. Adequate evidence for the formulation of a theory concerning mode of transmission of resistance from parent to offspring has not been presented.

8. Once resistance to any insecticide is developed by house flies, the development of resistance to other insecticides proceeds at an accelerated rate.

9. Resistance by other species of economic importance will probably be developed.

10. The several resistant strains of house flies do not behave in an analogous manner, and progeny from the same parent strain often have different responses to insecticides when tested in other laboratories. It is almost impossible, therefore, to compare directly results between laboratories.

11. The resistance of house flies is not due to the failure of the insecticide to penetrate the cuticle since resistance is evident when the insecticide is injected directly into the body cavity. Resistance is also apparent regardless of whether the insecticide is applied as a mist, dust, or residue.

Literature Cited

(1) Anonymous.
1949. Methoxychlor, a manual of technical data. E. I. du Pont de Nemours & Co., July 1.

(2) _____
1949. Controlling flies with BHC. Agr. Chem. 4(4): 30-32, 73, 75.

(3) _____
1949. Are insects rallying against DDT. Amer. Fruit Growers 69(2): 17, 39.

(4) _____
1950. How's your supply of chemicals? Do you have enough? Pest Control 18(9): 7.

(5) _____
1950. Pepper extract lets DDT kill super-flies. Farm Jour., March: 17.

(6) _____
1950. Switch chemicals in 1950 Dr. Decker says at Purdue conference. Pest Control 18(3): 12, 14, 16, 18, 20.

(7) _____
1950. Says tolerant fly structures same as others. Pest Control 18(4): 22.

(8) Ayars, James S.
1950. Flies thrive on DDT? Kill them with lindane. Successful Farming 48(7): 18, 104.

(9) Babers, Frank H.
1949. Development of insect resistance to insecticides. U.S. Bur. Ent. and Plant Quar. E-776, 31 pp. [Processed.]

(10) _____ and Pratt, Jr., John J.
1950. Studies on the resistance of insects to insecticides. I. Cholinesterase in house flies (Musca domestica L.) resistant to DDT. Physiol. Zool. 23: 58-63.

(11) Bailey, Stanley F.
1948. The peach twig borer. Calif. Agr. Expt. Sta. Bul. 708, 56 pp.

(12) Barber, George H., and Schmitt, John B.
1948. House flies resistant to DDT residual sprays. N. J. Agr. Expt. Sta. Bul. 742, 8 pp.

(13) Barber, George H., and Schmitt, John B.
1949. A line of house flies resistant to methoxychlor. Jour. Econ. Ent. 42: 844-845.

(14) _____ and Schmitt, John B.
1949. Further studies on resistance to DDT in the house fly. Jour. Econ. Ent. 42: 287-292.

(15) _____ Starnes, Ordway, and Starnes, Eleanor B.
1948. Resistance of houseflies to insecticides. Soap and Sanit. Chem. 24(11): 120, 121, 143.

(16) Berim, N. G., and Edelman, N. M.
1949. On some physiological factors which determine the resistance of insects to dichlorodiphenyl trichloroethane (DDT) and hexachlorocyclohexane. Dokl. Akad. Nauk. SSSR 67: 585-588 [English translation by R. Ericson.]

(17) Bertholf, J. H.
1950. DDT resistant mosquitoes in Broward County, Florida. Fla. Anti-Mosquito Assoc. Ann. Rpt. 21: 80-83. [Processed.]

(18) Bettini, Sergio.
1948. Contributo allo studio della resistenza all' azione del DDT nelle mosche domestiche. Riv. di Parassitol. 9: 137-142; Ist Super di Sanit. Rend. 11: 1131-1136.

(19) _____
1948. I nuovi insetticidi. Ist. Super di Sanit. Rend. 11: 821-840.

(20) _____ and Barachini, Benivieni.
1948. Primi resultati della lotta con l' octa-klor ed il Gammaesano contro le mosche domestiche resistenti al DDT. Ist. Super di Sanit. Rend. 11: 841-848.

(21) Bishopp, F. C., and Laake, E. W.
1921. Dispersion of flies by flight. Jour. Agr. Res. 21: 729-766.

(22) Blauvelt, W. E., and Hoffman, Julius R.
1948. Parathion aerosol for greenhouse pest control. N. Y. State Flower Growers Bul. 29, 6 pp.

(23) Blickle, Robert L., Capelle, Asher, and Morse, W. J.
1948. Insecticide resistant houseflies. Soap and Sanit. Chem. 24(8): 139, 141, 149.

(24) Bohart, R. M., and Murray, W. D.
1950. DDT resistance in Aedes nigromaculis larvae. Calif. Mosquito Control Assoc. Ann. Conf., Proc. and Papers: 18, 20-21. [Processed.]

(25) Bot, Jan.
1949. Bydrae tot die verklaring van die werking van die insektisides DDT, hexachlor, siklohexan, chlordane en toxaphene. Luctor et Emergo, Leiden. 135 pp.

(26) Bradley, George H., and Lyman, F. Earle.
1950. Discussion of five years use of DDT residuals against Anopheles quadrimaculatus. Jour. Natl. Malaria Soc. 9: 113-118.

(27) _____ and Lyman, F. Earle.
1950. Remarks on the present status of the malaria eradication program. Fla. Anti-Mosquito Assoc. Ann. Rpt. 21: 14-17. [Processed.]

(28) Brown, H. D., and Rogers, E. F.
1950. The insecticidal activity of 1,1-dianisyl-neopentane. Amer. Chem. Soc. Jour. 72: 1864-1865.

(29) Bruce, W. N.
1949. Latest report on fly control. Pest Control 17(6): 7, 28.

(30) _____
1950. Current report on housefly resistance to insecticides. Pest Control 18(4): 9-10, 19.

(31) _____
1950. Insect resistance to insecticides. Amer. Assoc. Econ. Ent. North Cent. States Br. Proc. 5: 33-34. [Processed.]

(32) _____
1950. "Resistant" Flies. Amer. Assoc. Econ. Ent. North Cent. States Br. Proc. 5: 13. [Processed.]

(33) _____ and Decker, G. C.
1950. House fly tolerance for insecticides. Soap and Sanit. Chem. 26(3): 122-125, 145-147.

(34) Bucher, G. E., Cameron, J. W. MacB., and Wilkes, A.
1948. Studies on the housefly (Musca domestica L.). II. The effects of low temperatures on laboratory reared puparia. Canad. Jour. Res., Sect. D, Zool. Sci. 26: 26-56.

(35) Busbey, R. L., Yust, H. R., and Fulton, Robert A.
1942. Toxicity of trichloroacetonitrile to the California red scale. Jour. Econ. Ent. 35: 452-453.

(36) Busvine, J. R.
1949. A test for DDT-resistant flies. Ceylon Jour. Med. Sci. 6: 181-183.

(37) Cain, Jr., Thomas L.
1950. Observations on DDT resistant species of mosquitoes found in Brevard County. Fla. Anti-Mosquito Assoc. Ann. Rpt. 21: 84-85. [Processed.]

(38) Cartwright, W. B., and Noble, W. B.
1947. Studies on biological races of the hessian fly. Jour. Agr. Res. 75: 147-153.

(39) Catalano, Giuseppe, and Mariani, Mario.
1948. Influenza dei solventi sull azione insetticida residua del DDT. Soc. Ital. di Biol. Sper. Bol. 24: 1042-1044.

(40) Cressman, A. W.
1944. Variations in the susceptibility of California red scale to oil sprays. Jour. Econ. Ent. 37: 851.

(41) Crow, James F.
1947. Differences in susceptibility to ether in the virilis group of Drosophila. Abstract in Genetics 33: 102, 1948.

(42) D'Alessandro, G., Catalano, G., Mariani, M., Scerrino, E., Smiraglia, Calogera, and Valguarnera, G.
1949. Sulla resistenza delle mosche al DDT. Sicilia Medica 6: 5-16. [English translation by R. Ericson.]

(43) _____ and Scerrino, Enrico.
1948. Sulla resistenza di un particolare ceppo di musca domestica al DDT residuo e nebulizzato. Soc. Ital. di Biol. Sper. Bol. 24: 1041-1042.

(44) _____ and Smiraglia, Calogera.
1949. Comportamento di stipiti di Musca domestica sensibili e resistenti al D.D.T. Di Fronte a esaclorocicloesano e Octa-Klor. Soc. Ital. di Biol. Sper. Bol. 25: 305-307.

(45) Davidovici, S., Levinson, Z., and Reuter, S.
1950. The toxicity of DDT-lanoline residues to flies and mosquitoes. Paper presented to Expert Committee on Malaria of World Health Organization (W.H.O./MAL/37, Mar. 20, 1950), 4 pp. [Processed.]

(46) Decker, George C.
1950. "Hell-Fire on fly resistance?" Pest Control 18(4): 11, 16.

(47) Deonier, C. C., Cain, Jr., T. L., and McDuffie, W. C.
1950. Aerial spray tests on adult salt-marsh mosquitoes resistant to DDT. Jour. Econ. Ent. 42: 506-510.

(48) _____ and Gilbert, I. H.
1950. Resistance of salt marsh mosquitoes to DDT and other insecticides. Mosquito News 10: 138-143.

(49) Ebeling, Walter.
1938. Host-determined morphological variations in Lecanium corni. Hilgardia 11: 611-631.

(50) Fay, Richard W., Baker, Walter C., and Grainger, Mary M.
1949. Laboratory studies on the resistance of Anopheles quadrimaculatus to DDT and other insecticides. Natl. Malaria Soc. Jour. 8: 137-146.

(51) Finch, F. H.
1948. Arsenic resistant cattle ticks and their reaction to gammexane. Past. Rev. 58: 537-538.

(52) Fjelddalen, Jack.
1949. Methoxychlor. DDT resistente fluer. Norsk. Landbruk. 15: 393-396.

(53) Fox, Allen S.
1947. Antigenic differences between mutant strains of Drosophila melanogaster. Abstract in Genetics 33: 104-105.

(54) Gahan, James B., Downs, Wilbur G., and Celis, Heliodoro.
1949. Control of Anopheles psuedopunctipennis in Mexico with DDT residual sprays applied in buildings. Part II. Amer. Jour. Hygiene 49: 285-289.

(55) _____ and Weir, John M.
1950. Houseflies resistant to benzene hexachloride. Science 111: 651-652.

(56) Garman, Philip.
1950. Parathion resistant red spiders. Jour. Econ. Ent. 43: 53-56.

(57) Gilbert, I. H., Wilson, H. G., and Coarsey, J. M.
1950. Control of house flies in barns with different insecticides. U. S. Bur. Ent. and Plant Quar. E-795, 11 pp. [Processed.]

(58) Goldstein, L.
1949. Contribution a l étude de la sensibilite Hereditaire au gaz carbonique chez la Drosophile. Bul. Biol. de la France et Belg. 83: 177-188.

(59) _____
1949. A mutant genoid in Drosophila sensitive to carbon dioxide. Nature (London) 164: 369.

(60) Haarer, A. E.
1950. Cattle ticks breed immunity to poisons. Leather Trades Rev. 95: 341.

(61) Hadjinicolau, J.
1948. Toxicity of DDT and gammexane sprays to house flies (Musca domestica L.). Greece Hypourg Hygienes Archeia Hygienis 6: 1. (Not seen.)

(62) Hansens, E. J.
1950. House fly control in dairy barns. Jour. Econ. Ent. 43: 852-858.

(63) _____ and Goddin, A. H.
1949. Reaction of certain fly strains to DDT and methoxychlor deposits. Jour. Econ. Ent. 42: 843-844.

(64) _____ Schmitt, John B., and Barber, George W.
1948. Resistance of house flies to residual applications of DDT in New Jersey. Jour. Econ. Ent. 41: 802-803.

(65) Harrison, Mary C.
1950. DDT resistant houseflies. Ann. Appl. Biol. (Assoc. Appl. Biol. Proc.) 37: 306-308.

(66) Hatfield, Ira.
1949. Fly control; chemicals and toxicities. Pest Control 17(6): 9-12, 14, 26.

(67) Hewlett, P. S., and Plackett, R. L.
1950. Statistical aspects of the independent joint action of poisons, particularly insecticides. II. Examination of data for agreement with the hypothesis. Ann. Appl. Biol. 37: 527-552.

(68) Jayewickreme, S. H.
1950. Resistance to the action of a mineral oil larvicide by Anopheles subpictus Grassi. Ceylon Jour. Sci. (B) 24: 117-121.

(69) Johnson, M. S., and Hill, A. J.
1948. Partial resistance of a strain of bed bugs to DDT residuals. Med. News Letter 12(1): 26-28.

(70) Kalina, Bernard Fram.
1950. Development and viability of Drosophila melanogaster on a medium containing DDT. Science 111: 39-40.

(71) Keiding, J., and Van Duers, H.
1949. DDT resistance in houseflies in Denmark. Nature [London] 163: 964.

(72) King, W. V.
1950. DDT-resistant house flies and mosquitoes. Jour. Econ. Ent. 43: 527-532.

(73) _____
1950. Studies of DDT-resistant mosquitoes in Florida. Fla. Anti-Mosquito Assoc. Ann. Rpt. 21: 77-79.

(74) _____ and Gahan, J. B.
1949. Failure of DDT to control house flies. Jour. Econ. Ent. 42: 405-409.

(75) Kjellander, Eric.
1949. De Resistenta Flugorna Nagra histioriska data och ulbredningsuppgifter. Lantmannen 33: 575-576, 588.

(76) _____
1950. Flugretrotningen Resistenta flugor. Ladugarden (June) 174-175.

(77) Knipling, E. F.
1949. DDT developments. Soap and Sanit. Chem. 25(7): 107, 109, 111, 131.

(78) _____
1949. DDT resistant house flies. Amer. Assoc. Econ. Ent. North Cent. States Br. Proc 4: 89-90.

(79) _____
1950. Insecticidal resistant flies and mosquitoes. Soap and Sanit. Chem. (Special Issue Official Proc. C.S.M.A.) 26: No. C.S.M.A. (1): 87-88.

(80) _____
1950. Mosquito problems and research in the Pacific Northwest. Fla. Anti-Mosquito Assoc. Ann. Rpt. 21: 12-13.

(81) Knipling, E. F.
1950. Fly resistance to DDT. Amer. Assoc. Econ. Ent. North Cent. States Br. Proc. 5: 13.

(82) Kono, Tatsuro.
1949. The influence of sublethal doses of poisonous gases on insects. I. Influence of sub-lethal doses on the susceptibility of the adult rice beetle, Calandra oryzae, to carbon disulfide. Botyu Kagaku 12: 19-23 [In Japanese. English summary, pp. 24-25.]

(83) La Face, Lidia
1948. La Mosca domestica, la sua importanza come vettore di malattie e la possible esistenza di piu razze nell' ambito della specie. Ist. Super di Sanit. Rend. 11: 1287-1345. Riv. di Parassitol. 9: 199-222.

(84) Läuger, P., Pulver, R., Montigel, C., Wiesmann, R., and Wild, H.
1946. Mechanism of intoxication of DDT insecticides in insects and warm-blooded animals. Geigy Co., New York. 24 pp.

(85) L'Heritier, Ph.
1948. Sensitivity to CO_2 in Drosophila. A review. Heredity 2: 325-348.

(86) _____
1948. Transmission de la sensibilite au CO_2 chez la Drosophile par voie hereditaire et voie d'inoculation. Abstract in Internatl. Cong. Genet. Proc. 8: 619-620.

(87) _____ and Plus, Nadine.
1950. Inactivation par les rayons X du virus responsable de la sensibilite au CO_2 chez la Drosophile. Paris Acad. des Sci. Compt. Rend. 231: 192-194.

(88) _____ and de Scoeux, F. Hugon.
1947. Transmission par greffe et injection de la sensibilite hereditaire au gas carbonique chez la Drosophile. Bul. Biol. de la France et Belg. 81: 70-90.

(89) _____ and Sigot, A.
1944. Contribution a l' étude de la sensibilite au CO_2 chez la Drosophile III. Suppression de la sensibilite par l'action de la temperature. Soc. Phys. Biol. Compt. Rend. 18: 2-3.

(90) _____ and Teissier, G.
1945. Transmission hereditaire de la sensibilite au gaz carbonique chez Drosophila melanogaster. Pub. Lab. l'ecole Norm. Super. Paris Biol. Fasc. 1: 35-74.

(91) Lindgren, D. L., and Dickson, R. C.
1942. Spray fumigation experiments on California red scale. Jour. Econ. Ent. 35: 827-829.

(92) Lindquist, Arthur W., Madden, A. H., and Schroeder, H. O.
1946. Effect of temperature on knockdown and kill of mosquitoes and bedbugs exposed to DDT. Kans. State Ent. Soc. Jour. 19: 13-15.

(93) _____ Roth, A. R., Yates, W. W., and Butts, Joseph S.
Use of radioactive tracers in studies of DDT in house flies. Jour. Econ. Ent. (In press.)

(94) _____ and Wilson, H. G.
1948. Development of a strain of houseflies resistant to DDT. Science 107: 276.

(95) _____ Wilson, H. G., Schroeder, H. O., and Madden, A. H.
1945. Effect of temperatures on knockdown and kill of houseflies exposed to DDT. Jour. Econ. Ent. 38: 261-264.

(96) Loiseleur, Jean.
1948. Sur la formation et proprietes des anticorps correspondant aux molecules organiques de faible poids moleculaire. Paris Acad. Sci. Compt. Rend. 226: 1861-1862.

(97) _____
1948. Sur la nature de la fonction anticorps correspondent aux molécules organiques de faible poids moléculaire. Paris Acad. Sci. Compt. Rend. 226: 1927-1928.

(98) March, Ralph B., and Lewallen, Lawrence L.
1950. A comparison of DDT-resistant and non-resistant house flies. Jour. Econ. Ent. 43: 721-722.

(99) _____ and Metcalf, R. L.
1949. Laboratory and field studies of DDT resistant houseflies in southern California. Calif. State Dept. Agr. Bul. 37: 93-101.

(100) _____ and Metcalf, R. L.
1949. Insecticide resistant houseflies. Development of resistance to organic insecticides other than DDT by houseflies in southern California. Co-op Poultryman 14(12): 17, 30. Nulaid News 27(9): 14-16. Calif. Agr. 3(11): 3.

(101) _____ and Metcalf, R. L.
1949. Development of resistance to organic insecticides other than DDT by houseflies. Jour. Econ. Ent. 42: 990.

(102) March, Ralph B., and Metcalf, R. L.
1950. Studies in California of insecticide-resistant flies. Soap and Sanit. Chem. (Special Issue Official Proc. C.S.M.A.) 26; No. C.S.M.A. (1): 80-83, Soap and Sanit. Chem. 26(7): 121, 123, 125, 139.

(103) _____ and Metcalf, R. L.
1950. Investigations of insecticide-resistant house flies in Southern California. Calif. Mosquito Control Assoc. Conf. Proc. and Papers 18: 17-20 [Processed.]

(104) Markos, Basil G., and Campbell, F. L.
1943. Effect of host plant on the susceptibility of the southern armyworm to calcium arsenate. Jour. Econ. Ent. 36: 662-665.

(105) Martin, H., and Wain, R. L.
1944. Insecticidal action of DDT. Nature [London] 154: 512-513.

(106) McDuffie, W. C.
1950. Experimental procedures for determining the occurrence of DDT resistant mosquitoes and suggestions for their control. Fla. Anti-Mosquito Assoc. Ann. Rpt. 21: 69-70.

(107) McGovran, E. R., and Gersdorff, W. A.
1945. The effect of fly food on resistance to insecticides containing DDT or pyrethrum. Soap and Sanit. Chem. 21(12): 165, 169.

(108) Melander, A. L.
1914. Can insects become resistant to sprays? Jour. Econ. Ent. 7: 167-172.

(109) Miller, A. C.
1949. Insecticide resistant houseflies. Amer. Assoc. Econ. Ent. North Cent. States Br. Proc. 4: 91-92.

(110) Missiroli, A.
1947. Riduzione o eradicagione degli anofeli. Riv. di Parassitol. 8: 141-169.

(111) Monro, H. A. U.
1949. Insect resistance to insecticides.--Parts I and II. Pest Control 17(9): 16, 18, 20, 42; 17(10): 22, 24, 26, 50.

(112) Morrison, H. E., Lauderdale, R. W., Crowell, H. H., and Mote, Don C.
1950. Space spraying for fly control in dairy barns. Jour. Econ. Ent. 43: 846-850.

(113) Mosna, Enzio.
1948. Su una caratteristica biologica del culex pipiens autogenicus di Latina. Ist. Super di Sanit. Rend. 11: 125-126.

(114) _____
1948. Culex pipiens autogenicus DDT-resistenti e loro controllo con octa-klor e esachlorocicloesano. Ist Super di Sanit. Rend. 11: 425-432.

(115) _____
1948. Culex pipiens autogenicus DDT-resistenti e loro controllo con octa-klor e esaclorocicloesano. Riv. di Parassitol. 9: 19-25.

(116) _____
1949. Octa-Klor, gammaesano, e toxaphene usati contro le mosche DDT-resistenti. Riv. di Parassitol. 10: 31-51; Ist. Super di Sanit. Rend. 12: 467-489.

(117) _____
1950. Il controllo con octa-klor delle mosche domestiche DDT-resistenti. Riv. di Parassitol. 11: 27-35.

(118) _____ and Alessandrini, M.
1950. La lotta antianofelica con DDT nella provincia di Latina Nel 1948 e 1949. Riv. di Parassitol. 11: 13-26.

(119) Munger, Francis.
1949. Rate of development of California red scales resistant and non resistant to hydrocyanic acid gas, as affected by temperature. Jour. Agr. Res. 78: 451-456.

(120) _____
1949. Effect of temperatures on California red scale. Calif. Citrog. 34: 464.

(121) Neiswander, C. R., and Morris, V. H.
1940. Introduction of selenium into plant tissues as a toxicant for insects and mites. Jour. Econ. Ent. 33: 517-525.

(122) _____ Rodriguez, J. G., and Neiswander, R. B.
1950. Natural and induced variations in two-spotted spider mite populations. Jour. Econ. Ent. 43: 633-636.

(123) Neri, Italia.
1948. Nota sulle differenze di sensibilita di Musca domestica dopo trattamento con DDT. Ist. Super di Sanit. Rend. 11: 1362-1385.

(124) Newman, J. F., Aziz, M. A., and Koshi, T.
1949. Studies in contact toxicity. Part 1. Resistance of successive generations of Culex fatigans Wied. to contact insecticides. Indian Acad. Sci. Proc. B. 30: 61-68.

(125) Painter, Reginald H.
1930. The biological strains of hessian fly. Jour. Econ. Ent. 23: 322-326.

(126) Palm, Charles E.
1949. Development and use of synthetic organic chemicals. Adv. Chem. Ser. 1: 218-222.

(127) Patrissi, T., Barbieri, V., and Bessler, C.
1949. Resistenza di Musca domestica al DDT ed impiego di altri insetticidi. Riv. Malariol. 28: 137-151.

(128) Perry, A. S., and Hoskins, W. M.
1950. The detoxification of DDT by resistant houseflies and inhibition of this process by piperonyl cyclonene. Science 111: 600-601.

(129) Pielou, D. P.
1950. Preparation of thin films of crystalline DDT and γ-hexachlorocyclohexane in celloidin. Science 112: 406-407.

(130) Pimentel, David, and Dewey, J. E.
1950. Laboratory tests with house flies and house fly larvae resistant to DDT. Jour. Econ. Ent. 43: 105.

(131) _____ Schwardt, H. H., Dewey, J. E., and Norton, L. B.
1950. Studies on insecticide resistant house flies in New York. Soap and Sanit. Chem. (Special Issue Official Proc. C.S.M.A.) 26: No. C.S.M.A. (2): 94-96.

(132) Plackett, R. L., and Hewlett, P. S.
1948. Statistical aspects of the independent joint action of poisons, particularly insecticides. I. The toxicity of a mixture of poisons. Ann. Appl. Biol. 35: 347-358.

(133) Plus, Nadine.
1950. L' invasion d'un organisme par un virus hereditaire; recherches quantitatives sur la vitesse d'acquisition de la sensibilite au CO_2 par les drosophiles. Expt. Cell. Res. 1: 217-233. Appendice mathématique by P. L'Heritier, pp. 230-233. [English summary.]

(134) Pratt, Jr., John J., and Babers, Frank H.
1950. Cross tolerances in resistant house flies. Science 112: 141-142.

(134a) Quarterman, Kenneth D.
1950. The status of fly resistance to insecticides in the Savannah area and its implications in the general problem of fly control. U.S. Pub. Health Serv. CDC Bul. 9(11): 3-7.

(135) Quayle, H. J.
1943. The increase in resistance in insects to insecticides. Jour. Econ. Ent. 36: 493-500.

(136) Raucourt, Marc.
1948. L' arséniate de plomb et le carpocapse. Rev. Hort. 120: 10.

(137) Reimschneider, Von R., and Rohrmann, B.
1950. Über die Zucht DFDT-resistenter Drosophila melanogaster M. Anz. f. Schädlingsk. 23(10): 148-149.

(138) Ricci, Marcello
1948. Sull' azione del DDT su Blatta orientalis. Riv. di Parassitol. 9: 143-167.

(139) Roth, A. R., and Mote, Don C.
1950. DDT-resistant flies in Oregon. Jour. Econ. Ent. 43: 937.

(140) Rubtsov, Ivan.
1948. The appearance of a form of house fly resistant to DDT as result of a 3-year application of insecticide. Priroda USSR (4): 62-63. [Translation by S. N. Monson.]

(141) Russell, Raymond.
1949. Control de moscas de establo. Agronomia (Peru) 14(58): 43-45.

(142) Sacca, Giuseppe.
1948. Sull' esistenza di mosche domestiche resistenti al DDT. Nota preliminare. Ist. Super di Sanit. Rend. 11: 123-124.

(143) ———
1948. Osservazioni su di una popolazione di mosche dopo trattamento con DDT e Octa-klor. Ist. Super di Sanit. Rend. 11: 1354-1361.

(144) Sacktor, Bertram.
1950. A comparison of the cytochrome oxidase activity of two strains of house flies. Jour. Econ. Ent. 43: 832-838.

(145) Schaerffenberg, Bruno.
1949. Über die Erntrittsstellen der Kontakgifte und die Ursachen der DDT-Resistenz der Maikäferlarvae. Ztschr. f. Pflanzenkrank. 56: 37-39.

(146) Shepard, Harold H.
1939. The chemistry and toxicology of insecticides. 383 pp. Minneapolis.

(147) Smith, Floyd F., and Fulton, R. A.
1949. Red spider resistance to aerosols and sprays. Florists Exch. Hort. Trade World 113 (23): 15, 49-51.

(148) _____ and Fulton, R. A.
1949. Spider mites resistance to aerosol sprays investigated. South. Florists and Nurseryman 62(37): 16-17, 107-110.

(149) _____ and Fulton, R. A.
1949. Strains of red spiders are resistant to some insecticides. Florists Rev. 105(2718): 23-25.

(150) _____ Fulton, R. A., and Lung, P. H.
1948. Recent developments in the control of greenhouse pests by liquefied-gas aerosols. Jour. Econ. Ent. 41: 624-631.

(151) Smith, H. S.
1941. The segregation of insect populations into races and its significance in applied entomology. Jour. Econ. Ent. 34: 1-13.

(152) Smith, John B.
1897. The influence of environment on the life history of insects. Gard. and Forest 10: 334.

(153) Sternberg, James, and Kearns, C. W.
1950. Degradation of DDT by resistant and susceptible strains of house flies. Ent. Soc. Amer. Ann. 43: 444-458.

(154) _____ Kearns, C. W., and Bruce, W. N.
1950. Absorption and metabolism of DDT by resistant and susceptible houseflies. Jour. Econ. Ent. 43: 214-219.

(155) Summers, Francis M.
1949. Resistance to basic lead arsenate by the peach twig borer within a small area in central California. Jour. Econ. Ent. 42: 22-24.

(156) Swingle, M. C.
1939. The effect of previous diet on the toxic action of lead arsenate to a leaf feeding insect. Jour. Econ. Ent. 32: 884.

(157) Thorburn, J. A.
1947. The control of ectoparasitic infestations of farm stock with "Gammexane" with special reference to the arsenic-resistant blue tick. Empire Jour. Expt. Agr. 15: 42-50.

(158) Thorpe, W. H.
1930. Biological races in insects and allied groups. Cambridge Phil. Soc. Biol. Rev. 5: 177-212.

(159) Turner, Neely.
1950. New laboratory techniques speed pesticide experiments. Frontiers Plant Sci. 2(2): 4-5.

(160) Upholt, W. M.
1950. Significance to the insecticide industry of fly resistance. Soap and Sanit. Chem. (Special Issue Official Proc. C.S.M.A.) 26: No. C.S.M.A. (2): 69-71.

(161) Verolini, Felice.
1948. Sul meccanismo di azione del DDT sulle larve di culicidi. Ist. Super di Sanit. Rend. 11: 442-449.

(162) Wachs, Herman.
1947. Synergistic insecticides. Science 105: 530-531.

(163) Walsh, Benjamin D.
1865. On phytophagic varieties and pytophagic species. Ent. Soc. Phila. Proc. 5: 194-216.

(164) Whitnall, A. B.
1949. Blue ticks' resistance to B.H.C. proved. Primary Prod. (Cape Town) 24: 19.

(165) _____ and Bradford, B.
1947 An arsenic resistant tick and its control with "Gammexane" dips. Bul. Ent. Res. 38: 353-372.

(166) _____ and Bradford, B.
1949. An arsenic resistant tick and its control with "Gammexane" dips. Part 2. Bul. Ent. Res. 40: 207-226.

(167) _____ Thorburn, J. A., Whitehead, G. B., McHardy, W. M., and Meerholz, F.
1949. A tick (Boophilus decoloratus) resistant to γ-benzene hexachloride. Nature [London] 164: 956-957.

(168) Wiesmann, R. von.
1947. Untersuchungen uber das physiologische Verhalten von Musca domestica L. verschiedener Provenienzen. Schweiz. Ent. Gesell. Mitt. 20: 484-504.

(169) Wilson, Charles S.
1949. Piperonyl butoxide, piperonyl cyclonene, and pyrethrum applied to selected parts of individual flies. Jour. Econ. Ent. 42: 423-428.

(170) Wilson, H. G., and Gahan, J. B.
1948. Susceptibility of DDT-resistant houseflies to other insecticidal sprays. Science 107: 276-277.

(171) Yust, Harold R., Nelson, Howard D., and Busbey, R. L.
1943. Influence of inbreeding on the susceptibility of the California red scale to HCN. Jour. Econ. Ent. 36: 352-353.